tyrosine phosphatase activity at membranes, and abnormal smooth muscle cell proliferation, suggesting that PrxII does play an active role in H_2O_2 signaling (Choi et al., 2005). Future studies with mice that lack either or both PrxI and PrxII may help to illuminate how these two enzymes coordinate their activity to optimize H_2O_2 signaling at particular receptor kinases, while preventing the toxic effects of H_2O_2.

ACKNOWLEDGMENTS

M.B.T. is the recipient of a program grant from La Ligue Contre le Cancer (LNCC).

REFERENCES

Choi, M.H., Lee, I.K., Kim, G.W., Kim, B.U., Han, Y.H., Yu, D.Y., Park, H.S., Kim, K.Y., Lee, J.S., Choi, C., et al. (2005). Nature *435*, 347–353.

D'Autréaux, B., and Toledano, M.B. (2007). Nat. Rev. Mol. Cell Biol. *8*, 813–824.

Lambeth, J.D. (2004). Nat. Rev. Immunol. *4*, 181–189.

Meng, T.C., Fukada, T., and Tonks, N.K. (2002). Mol. Cell *9*, 387–399.

Tonks, N.K. (2006). Nat. Rev. Mol. Cell Biol. *7*, 833–846.

Ushio-Fukai, M. (2006). Sci. STKE *2006*, re8.

Woo, H.A., Yim, S.H., Shin, D.H., Kang, D., Yu, D.-Y., and Rhee, S.G. (2010). Cell, this issue.

Wood, Z.A., Poole, L.B., and Karplus, P.A. (2003). Science *300*, 650–653.

Rac in the Act of Forgetting

Ronald L. Davis[1,*]
[1]Department of Neuroscience, The Scripps Research Institute Florida, Jupiter, FL 33458, USA
*Correspondence: rdavis@scripps.edu
DOI 10.1016/j.cell.2010.02.004

Forgetting has been thought to occur as a result of the natural decay of the neuronal changes induced by learning or because of interference from other cognitive functions. In this issue, Shuai et al. (2010) find that the small G protein Rac may function as a switch for remembering versus forgetting.

Forgetting is undervalued. Although our forgetful nature is often a source of irritation, our lives would be chaos without forgetting given the mass of information that impinges on us daily. It is reasonable to think that our forgetfulness is passive, caused by the simple reversibility of molecular and cellular processes engaged when memories are first laid down in our brains. Alternatively, forgetting could be active, controlled by molecular and cellular mechanisms that erase unused or unwanted memories.

Research and thought in psychology over most of the 20th century have viewed forgetting in these two lights—as due to passive decay or to active interference from other mental activities that may rub out existing memories (Wixted, 2004; Jonides et al., 2008). In this issue, Shuai et al. (2010) have crystallized the notion that the brain mechanisms for remembering are balanced by a mechanism for active forgetting. Moreover, they have ushered in a completely new line of research—the cell biology of active forgetting—by identifying the G protein Rac as critical for active forgetting.

The investigators take advantage of the fruit fly *Drosophila*, pairing a robust learning assay with sophisticated genetic tools for restricting the expression of developmentally deleterious transgenes of *rac* (McGuire et al., 2004) to only the adult phase. Flies are particularly good at learning about odors that are paired with a mild electric shock (Davis, 2005). In the initial experiments, transgenic flies that express a dominant-negative Rac, *Drac1(N17)*, in all adult neurons are found to have a more persistent memory of the odor:shock pairing compared to controls. In contrast, expression of a constitutively active form of Rac, *Drac1(V12)*, has the opposite effect. It accelerates memory decay after learning. Other experiments show that these effects are due to changes in the stability of memory rather than the level of learning (acquisition) about the odor:shock association. Thus, Rac acts a rheostat for the stability of memory that can be dialed up or down, rather than functioning as a filter for memory acquisition.

Rac is a member of the Rho family of GTPases with roles in multiple signaling pathways that control transcription, cytoskeletal organization, vesicle trafficking, and cellular proliferation (Bustelo et al., 2007). This prompts the question, which of the many signaling pathways that utilize Rac underlie its memory functions? Cofilin is a potent actin-depolymerizing molecule that is inhibited by phosphorylation through the sequential activation of Rac, p21-activated kinase (PAK), and LIM-domain-containing protein kinase (LIMK) (Figure 1). Flies expressing a dephosphorylated and thus persistently active mutant of cofilin, the product of the *twinstar* gene *(tsr)*, exhibit enhanced memory similar to flies expressing dominant-negative Rac. This suggests that actin/cytoskeletal dynamics are at the heart of the function of Rac in memory.

Where in the brain does Rac function for memory? Olfactory memory in flies and other insects largely resides in the mushroom body (MB) neurons, which are third order neurons in the olfactory pathway and perhaps similar in function to neurons located in the primary olfactory cortex of vertebrates (Davis, 2004). When dominant-negative Rac is expressed only in the adult MB neurons, the resulting memory enhancement is the same as that observed with pan-neuronal expression, whereas expression in several different brain regions has no effect. Further attempts to map the memory enhancement to one of the three classes of MB neurons (α/β, α'/β', or γ) failed, suggesting that the enhancement requires pan-MB expression.

Learning new information after a prior learning event can interfere with the memory of the information learned first. The authors test memory-enhancing effect of dominant-negative Rac expression in such an interference paradigm in which flies are trained with one odor paired with electric shock and then trained with a second odor 90 min later. Control flies exhibited retroactive interference. The interference training reduces the memory of the initial odor when tested 3 hr later. Remarkably, flies expressing the dominant-negative Rac transgene show no interference—the stronger memory of the first learning event in these flies made them resistant to the interference caused by the second event.

The authors also test the role of Rac in situations when a memory becomes less relevant in an experimental paradigm known as reversal learning. The authors trained flies with one odor paired with electric shock (conditioned stimulus+, CS+) and exposed them to a second odor unpaired with electric shock (conditioned stimulus–, CS–). They were then immediately retrained but after switching the two odors, such that the CS– in the first training became the CS+ in the second. Flies normally display a stronger memory for the most recently trained odor, a phenomenon referred to as a recency effect. However, flies expressing dominant-negative Rac exhibit a weakened recency effect: the stronger memory of the first CS+/unconditioned stimulus pairing in these flies partially blocks learning of the second pairing. Moreover, flies expressing the constitutively active Rac transgene exhibit a stronger recency effect. The more rapid erasure of the memory of the first pairing by activated Rac enhances learning of the second pairing.

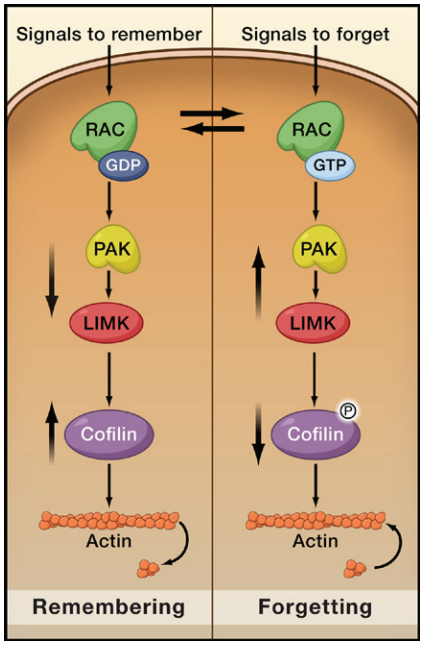

Figure 1. Rac Activity and the Persistence of Memory

The activity of Rac shifts the balance between remembering and forgetting. In the activated state (GTP-bound), Rac activates p21-activated kinase (PAK) and LIM-domain-containing protein kinase (LIMK) (arrow on the right indicates increased activity) leading to the phosphorylation and inactivation (decreased activity) of the actin-depolymerizing agent, cofilin. This shifts the actin equilibrium from the monomeric state to the filamentous state. Expression of a constitutively active Rac transgene in the fruit fly *Drosophila* produces more rapid forgetting after odor learning. Cofilin remains in the dephosphorylated and active form when Rac is in its inactive state (GDP-bound). The expression of a dominant-negative Rac in *Drosophila* produces enhanced remembering after odor learning.

Rac thus appears to be a rheostat that regulates the activity of cofilin, dynamics of actin polymerization, and forgetting or remembering (Figure 1). To probe the biochemical status of Rac during learning, activated Rac was measured in the heads of flies trained several times or in flies subjected to reversal learning. Intriguingly, the amount of activated Rac decreases with an increasing number of training trials, suggesting that multiple training trials suppress the Rac-based forgetting mechanism to facilitate remembering. Reversal learning, which erases memory of the first learning event, increased the level of activated Rac. The increased Rac activity should decrease cofilin activity, increase actin polymerization, and favor forgetting. It appears that repetitive training—a signal to remember—keeps Rac in check, whereas reversal learning—a signal to forget—potentiates Rac activity.

A major question concerns the identity of the upstream signaling events that lead to the balancing act that Rac may play (Figure 1). These are unknown, although the memory-enhancing effect of dominant-negative Rac is unaffected in the learning mutant, *rutabaga*, which encodes an adenylyl cyclase that functions as a coincidence detector for memory acquisition (Tomchik and Davis, 2009). Thus, the memory functions of Rac appear to be independent of cyclic AMP signaling at least through this important cyclase. Future experiments are needed to determine both upstream signaling events as well as how Rac activity alters actin dynamics.

What is the relationship between actin dynamics, remembering, and forgetting? It is generally believed that structural changes in synaptic morphology, mediated by rearrangements in the actin cytoskeleton, underlie the stabilization of memories after learning (Lamprecht and LeDoux, 2004). The *Drosophila* results on the role of Rac in forgetting are consistent with some observations made using the mouse. Mutant mice lacking LIMK1, which increases cofilin as expected in the dominant-negative Rac-expressing flies, have enhanced hippocampal long-term potentiation and enhanced memory of fear conditioning (Meng et al., 2002). Moreover, these mice exhibit reduced performance after reversal learning of the spatial water maze, similar to the reduction in performance after reversal odor learning seen in flies expressing dominant-negative Rac. Nevertheless, the results from *Drosophila* may come to most researchers as a surprise, given that a large body of experimental results suggest that long-term potentiation in the mammalian hippocampus and memory consolidation are promoted through gains in filamentous actin (Lamprecht

and LeDoux, 2004), whereas the Rac dominant-negative transgene, which enhances memory in flies, is expected to produce a decrease in filamentous actin (Figure 1). It may be that an incomplete understanding of the signaling systems involved underlies the discordance in these results. Alternatively, the different stages of memory—acquisition, consolidation, and forgetting—may require distinct cytoskeletal arrangements.

REFERENCES

Bustelo, X.R., Sauzeau, V., and Berenjeno, I.M. (2007). Bioessays *29*, 356–370.

Davis, R.L. (2004). Neuron *44*, 31–48.

Davis, R.L. (2005). Annu. Rev. Neurosci. *28*, 275–302.

Jonides, J., Lewis, R.L., Nee, D.E., Lustig, C.A., Berman, M.G., and Moore, K.S. (2008). Annu. Rev. Psychol. *59*, 193–224.

Lamprecht, R., and LeDoux, J. (2004). Nat. Rev. Neurosci. *5*, 45–54.

McGuire, S.E., Roman, G., and Davis, R.L. (2004). Trends Genet. *20*, 384–391.

Meng, Y., Zhang, Y., Tregoubov, V., Janus, C., Cruz, L., Jackson, M., Lu, W.Y., MacDonald, J.F., Wang, J.Y., Falls, D.L., and Jia, Z. (2002). Neuron *35*, 431-441.

Shuai, Y., Lu, B., Hu, Y., Wang, L., Sun, K., and Zhong, Y. (2010). Cell, this issue.

Tomchik, S., and Davis, R.L. (2009). Neuron *64*, 510–521.

Wixted, J.T. (2004). Annu. Rev. Psychol. *55*, 235–269.

Circadian Cell-Cycle Progression: Cracking Open the Gate

Jörg Morf[1,*] and Ueli Schibler[1,*]

[1]Department of Molecular Biology and NCCR Frontiers in Genetics, Sciences III 30, Quai Ernest Ansermet, CH-1211 Geneva-4, Switzerland

*Correspondence: joerg.morf@unige.ch (J.M.), ueli.schibler@unige.ch (U.S.)

DOI 10.1016/j.cell.2010.02.002

In cyanobacteria cell division is intimately linked with the circadian cycle. Dong et al. (2010) now identify components of the circadian clock that regulate the formation of the midcell ring for cytokinesis, revealing a critical link between the circadian cycle and the control of cell division.

Most light-sensitive organisms execute at least two fundamental processes that exhibit periodicity—cell-cycle progression and circadian physiology. Although the period length of endogenous circadian oscillators is approximately 24 hr, the length of the cell division cycle varies greatly among species. Although considerable progress has been made in uncovering the mechanisms and pathways controlling both of these cyclic processes, research aimed at understanding their interconnection is still in its infancy. In this issue, Dong et al. (2010) shed light on how circadian clock components impose restraints on the timing of cell division in the cyanobacterium *Synechococcus elongatus*.

We have recently witnessed groundbreaking progress in understanding the clockwork circuitry of cyanobacteria, including the reconstitution of a functional phosphorylation clock in vitro with only three proteins (KaiC, KaiA, KaiB) and adenosine triphosphate (Nakajima et al., 2005). KaiC is the only protein in the trio with known enzymatic activities, in that it can function as an autokinase, an autophosphatase, and an ATPase. The cyanobacteria "test tube-oscillator" still exhibits several key features of circadian clocks: it runs with a period length close to 24 hr, it is temperature compensated, and it can be synchronized (by the addition of ATP or KaiC subunit exchange).

The possibility of assembling a working clock from a small number of components has afforded detailed structure-function predictions and the examination of their validity by relatively straightforward biochemical experiments (Markson and O'Shea, 2009). Furthermore, in cyanobacteria the adaptive advantage of possessing an endogenous timing system has been clearly demonstrated. For example, in cocultures of cyanobacterial strains with or without functional circadian clocks, the strain with a functional clock outcompetes arrhythmic *kaiC* mutant strains when grown under light:dark cycles with a periodicity of 24 hr. However, this growth advantage disappears in cyanobacteria exposed to constant light (Woelfle et al., 2004). Moreover, if two mutant strains of cyanobacteria with circadian oscillators producing different period lengths are cocultured in light:dark cycles of different durations, the one with a resonating clock outgrows the one with the discordant oscillator after a few generations. These differences in fitness reflect interactions of endogenous clocks with external timing cues, rather than intrinsically different growth rates. In fact, when grown individually the examined wild-type and *kaiC* mutant stains proliferate at similar rates—approximately two doublings of the population per day in constant light and one doubling per day in circadian light:dark cycles (Woelfle et al., 2004).